36978

FORMULA: various

M.W.: various

ASBESTOS FIBERS

METHOD: 7402

ISSUED: 8/15/87

OSHA: 0.2 asbestos fibers (>5 µm long/mL)
NIOSH: 0.1 asbestos f/mL [1]
ACGIH: 0.2 crocidolite; 0.5 amosite; 2 chrysotile and other asbestos, f/mL

PROPERTIES: solid, fibrous

SYNONYMS: actinolite asbestos [CAS #13768-00-8], grunerite asbestos (amosite) [CAS #12172-73-5], anthophyllite asbestos [CAS #17068-78-9], chrysotile asbestos [CAS #12001-29-5], crocidolite asbestos [CAS #12001-28-4], tremolite asbestos [CAS #14567-73-8].

SAMPLING

SAMPLER: FILTER (0.8-to 1.2-µm cellulose ester membrane, 25-mm diameter; conductive cassette)

FLOW RATE*: 0.5 to 16 L/min (step 4)

VOL-MIN*: 400 L @ 0.1 fiber/mL (step 4)
-MAX*: (step 4)
*Adjust for 100 to 1300 fibers/mm² (step 4)

SHIPMENT: routine (securely packed to reduce shock)

SAMPLE STABILITY: stable

FIELD BLANKS: 10% (≥2) of samples

ACCURACY

RANGE STUDIED: 80 to 100 fibers counted

BIAS: not determined

OVERALL PRECISION (s_r): see EVALUATION OF METHOD

MEASUREMENT

TECHNIQUE: MICROSCOPY, TRANSMISSION ELECTRON (TEM)

ANALYTE: asbestos fibers

SAMPLE PREPARATION: modified Jaffe wick

EQUIPMENT: transmission electron microscope; energy dispersive X-ray system (EDS) analyzer

CALIBRATION: qualitative electron diffraction; calibration of TEM magnification and EDS system

RANGE: 100 to 1300 fibers/mm² filter area [2]

ESTIMATED LOD: 1 confirmed asbestos fiber above 95% of expected mean blank value

PRECISION: 0.28 when 65% of fibers are asbestos; 0.20 when adjusted fiber count is applied to PCM count [3].

APPLICABILITY: The working range is 0.04 to 0.5 fiber/mL for a 1-m³ air sample. The method measures asbestos fibers of the smallest diameter (<0.05 µm) but allows comparison of fiber counts to be made with phase contrast light microscopic (PCM) data when fiber diameters are rigidly defined.

INTERFERENCES: Non-asbestiform amphiboles may interfere in the TEM analysis if the individual particles have aspect ratios greater than 3:1. These interferences can only be eliminated by quantitative zone axis electron diffraction analysis. High concentrations of background dust interfere with fiber identification.

OTHER METHODS: NIOSH Method 7400 (dated 8/15/87) (phase-contrast microscopy) designed for use with this method.

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REAGENTS:

1. Acetone. See SPECIAL PRECAUTIONS.

EQUIPMENT:

1. Sampler: field monitor, 25-mm, three-piece cassette with ca. 50-mm electrically-conductive extension cowl, cellulose ester membrane filter, 0.8- to 1.2-µm pore size, and backup pad.
   NOTE 1: Analyze representative filters for fiber background before use. Discard the filter lot if mean count is >5 fibers/100 fields. These are defined as laboratory blanks.
   NOTE 2: Use an electrically-conductive extension cowl to reduce electrostatic effects on fiber sampling and during sample shipment. Ground the cowl when possible during sampling.
2. Personal sampling pump, $\geq$0.5 L/min (see step 4 for flow rate), with flexible connecting tubing.
3. Microscope, transmission electron, operated at 100 kV, with electron diffraction and energy-dispersive X-ray capabilities, and having a fluorescent screen with inscribed or overlaid calibrated scale (Step 15).
   NOTE: The scale is most efficient if it consists of a series of lines inscribed on the screen or partial circles every 2 cm distant from the center.
4. Diffraction grating replica with known number of lines/mm.
5. Slides, glass, pre-cleaned, 25- x 75-mm.
6. Knife, #10 surgical steel, curved-blade.
7. Tweezers.
8. Grids, 200-mesh TEM copper, carbon-coated.
9. Petri dishes, 15-mm depth. The top and bottom of the petri dish must fit snugly together. To assure a tight fit, grind the top and bottom pieces together with an abrasive such as carborundum to produce a ground-glass contact surface.
10. Foam, clean polyurethane, spongy, 12-mm thick.
11. Low-temperature oxygen plasma asher.
12. Filters, Whatman No. 1 qualitative paper or equivalent, or lens paper.
13. Vacuum evaporator.
14. Cork borer, No. 5 (8-mm).
15. Pen, waterproof, marking.
16. Reinforcement, page, gummed.
17. Asbestos standard bulk materials for reference.
18. Carbon rods, sharpened to 1 mm x 8 mm.
19. Microscope, light, phase contrast (PCM), with Walton-Beckett graticule (see method 7400).
20. Grounding wire, 22-gauge, multi-strand.

SPECIAL PRECAUTIONS: Acetone is extremely flammable (flash point = 0 °F). Take precautions not to ignite it. Heating of >1 mL acetone must be done in a fume hood using a flameless, spark-free heat source.

SAMPLING:

1. Calibrate each personal sampling pump with a representative sampler in line [4].
2. For personal sampling, fasten sampler to worker's lapel near worker's mouth. Remove the top cover from end of the cowl extension (open face) and orient sampler face down. Wrap joint between extender and monitor body with shrink tape to prevent air leaks. Where possible, especially at low %RH, attach sampler to electrical ground to reduce electrostatic effects during sampling.

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3. Submit at least two field blanks (or 10% of the total samples, whichever is greater) for each set of samples. Remove top covers from the field blank cassettes and store top covers and cassettes in a clean area (e.g., closed bag or box) during sampling. Replace top covers when sampling is completed.
4. Sample at 0.5 L/min or greater [5]. Adjust sampling rate, Q (L/min), and time, t (min), to produce fiber density, E, of 100 to 1300 fibers/mm² [$3.85 \cdot 10^4$ to $5 \cdot 10^5$ fibers per 25-mm filter with effective collection area (A_c = 385 mm²)] for optimum accuracy. Do not exceed ca. 0.5 mg total dust loading on the filter. These variables are related to the action level (one-half the current standard), L (fibers/mL), of the fibrous aerosol being sampled by:

$$t = \frac{A_c \cdot E}{Q \cdot L \cdot 10^3}, \text{ min.}$$

NOTE: The purpose of adjusting sampling times is to obtain optimum fiber loading on the filter. A sampling rate of 1 to 4 L/min for 8 hrs (700 to 2800 L) is appropriate in non-dusty atmospheres containing ca. 0.1 fiber/mL. Dusty atmospheres require smaller sample volumes (≤400 L) to obtain countable samples. In such cases take short, consecutive samples and average the results over the total collection time. For documenting episodic exposures, use high rates (7 to 16 L/min) over shorter sampling times. In relatively clean atmospheres, where targeted fiber concentrations are much less than 0.1 fiber/mL, use larger sample volumes (3000 to 10000 L) to achieve quantifiable loadings. Take care, however, not to overload the filter with background dust [5].

5. At the end of sampling, replace top cover and small end caps.
6. Ship samples upright with conductive cowl attached in a rigid container with packing material to prevent jostling or damage.

NOTE: Do not use untreated polystyrene foam in the shipping container because electrostatic forces may cause fiber loss from sample filter.

SAMPLE PREPARATION:

7. Remove a circular section from any quadrant of each sample and blank filter using a cork borer [6].
8. Affix the circular filter section to a clean glass slide with a gummed page reinforcement. Label the slide with a waterproof marking pen.

NOTE: Up to eight filter sections may be attached to the same slide.

9. Place the slide in a petri dish which contains several paper filters soaked with 2 to 3 mL acetone. Cover the dish. Wait 2 to 4 min for the sample filter(s) to fuse and clear.

NOTE: The "hot block" clearing technique may be used instead of steps 8 and 9 [7,8].

10. Place the slide containing the collapsed filters into a low-temperature plasma asher. Etch at 100 °C for ca. 2 min at an O_2 pressure of 130 Pa (1 mm Hg) [9].

NOTE: Plasma ashers may vary. Determine optimum etching time (ca. 1/2 the time needed to completely ash a filter) on blank filters before etching samples.

11. Transfer the slide to a rotating stage inside the bell jar of a vacuum evaporator. Evaporate a 1- by 5-mm section of a graphite rod onto the cleared filter(s). Remove the slide to a clean, dry, covered petri dish [6].
12. Prepare a second petri dish as a Jaffe wick washer with the wicking substrate prepared from filter or lens paper placed on top of a 12-mm thick disk of clean, spongy polyurethane foam [10]. Cut a V-notch on the edge of the foam and filter paper. Use the V-notch as a reservoir for adding solvent.

NOTE: The wicking substrate should be thin enough to fit into the petri dish without touching the lid.

13. Place the carbon-coated TEM grids face up on the filter or lens paper. Label the grids by marking with a pencil on the filter paper or by putting registration marks on the petri dish halves and marking with a waterproof marker on the dish lid. In a fume hood, fill the dish with acetone until the wicking substrate is saturated.
    NOTE: The level of acetone should be just high enough to saturate the filter paper without creating puddles.
14. Remove about a quarter section of the carbon-coated filter from the glass slide using a surgical knife and tweezers. Carefully place the excised filter, carbon side down, on the appropriately-labeled grid in the acetone-saturated petri dish. When all filter sections have been transferred, slowly add more solvent to the wedge-shaped trough to bring the acetone level up to the highest possible level without disturbing the sample preparations. Cover the petri dish. Elevate one side of the petri dish by placing a slide under it (allowing drops of condensed acetone to form near the edge rather than in the center where they would drip onto the grid preparation).

CALIBRATION AND QUALITY CONTROL:

15. Determine the TEM magnification on the fluorescent screen:
    a. Define a field of view on the fluorescent screen either by markings or physical boundaries.
       NOTE: The field of view must be measurable or previously inscribed with a scale or concentric circles (all scales should be metric) [10].
    b. Insert a diffraction grating replica into the specimen holder and place into the microscope. Orient the replica so that the grating lines fall perpendicular to the scale on the TEM fluorescent screen. Ensure that goniometer stage tilt is zero.
    c. Adjust microscope magnification to 10,000X. Measure the distance (mm) between the same relative positions (e.g., between left edges) of two widely-separated lines on the grating replica. Count the number of spaces between the lines.
       NOTE: On most microscopes the magnification is substantially constant only within the central 8 to 10 cm diameter region of the fluorescent screen.
    d. Calculate the true magnification (M) on the fluorescent screen:

$$M = \frac{X \cdot G}{Y}$$

       where: X = total distance (mm) between the two grating lines;
       G = calibration constant of the grating replica (lines/mm);
       Y = number of grating replica spaces counted
    e. After calibration, note the apparent sizes of 0.25 and 3.0 µm on the fluorescent screen. (These dimensions are essentially the diameter boundary limits for counting asbestos fibers by phase contrast microscopy.)
16. Measure 20 grid openings at random on a 200-mesh copper grid by placing a grid on a glass slide and examining it under the PCM. Use the Walton-Beckett graticule to measure the grid opening diameters. Calculate an average graticule field diameter from the data and use this number to calculate the graticule field area for an average grid opening.
    NOTE: A grid opening is considered as one graticule field.
17. Obtain reference selected area electron diffraction (SAED) or microdiffraction patterns from standard asbestos materials prepared for TEM analysis.
    NOTE: This is a visual reference technique. No quantitative SAED analysis is required [10]. Microdiffraction may produce clearer patterns on very small fibers or fibers partially obscurred by other material.
    a. Set the specimen holder at zero tilt.

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b. Center a fiber, focus, and center the smallest field-limiting aperture on the fiber. Use a 20-cm camera length and 10X binocular head. Obtain a diffraction pattern. Photograph each distinct 're pattern and keep the photo for comparison to unknowns.
   NOTE: Not all fibers wil. present diffraction patterns. The objective lens current may need adjustment to give optimum pattern visibility. There are many more amphiboles which give diffraction patterns similar to the analytes named on p.7402-1. Some, but not all, of these can be eliminated by chemical separations. Also, some non-amphiboles (e.g., pyroxenes, some talc fibers) may interfere.

18. Acquire energy-dispersive X-ray (EDX) spectra on approximately 5 fibers having diameters between 0.25 and 0.5 µm of each asbestos variety obtained from standard reference materials [10].
   NOTE: The sample may require tilting to obtain adequate signal. Use same tilt angle for all spectra.
   a. Prepare TEM grids of all asbestos varieties.
   b. Use acquisition times (at least 100 sec) sufficient to show a silicon peak at least 75% of the monitor screen height at a vertical scale of ≥500 counts per channel.
   c. Estimate the elemental peak heights visually as follows:
      (1) Normalize all peaks to silicon (assigned an arbitrary value of 10).
      (2) Visually interpret all other peaks present and assign values relative to the silicon peak.
      (3) Determine an elemental profile for the fiber using the elements Na, Mg, Si, Ca, and Fe. Example: 0-4-10-3-<1 [10].
      NOTE: In fibers other than asbestos, determination of Al, K, Ti, S, P, and F may also be required for fiber characterization.
      (4) Determine a typical range of profiles for each asbestos variety and record the profiles for comparison to unknowns.

MEASUREMENT:

19. Perform a diffraction pattern inspection on all sample fibers counted under the TEM, using the procedures given in step 17. Assign the diffraction pattern to one of the following structures:
   a. chrysotile;
   b. amphibole;
   c. ambiguous;
   d. none.
   NOTE: There are some crystalline substances which exhibit diffraction patterns similar to those of asbestos fibers. Many of these, (brucite, halloysite, etc.) can be eliminated from consideration by chemistry. There are, however, several minerals (e.g., pyroxenes, massive amphiboles, and talc fibers) which are chemically similar to asbestos and can be considered interferences. The presence of these substances may warrant the use of more powerful diffraction pattern analysis before positive identification can be made. If interferences are suspected, morphology can play an important role in making positive identification.

20. Obtain EDX spectra in either the TEM or STEM modes from fibers on field samples using the procedure of step 18. Using the diffraction pattern and EDX spectrum, classify the fiber:
   a. For a chrysotile structure, obtain EDX spectra on the first five fibers; one out of ten thereafter. Label the range profiles from 0-5-10-0-0 to 0-10-10-0-0 as "chrysotile."
   b. For an amphibole structure, obtain EDX spectra on the first 10 fibers; one out of ten thereafter. Label profiles ca. 0-2-10-0-7 as "possible amosite"; profiles ca. 1-1-10-0-6 as "possible crocidolite"; profiles ca. 0-4-10-3-<1 as "possible tremolite"; and profiles ca. 0-3-10-0-1 as "possible anthophyllite."

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Subtask 2 - Air Sampling

All air samples will be collected in accordance with NIOSH Method 7400 (copy attached).

The monitors will be distributed around the site as follows. One monitor will be placed on-site near exposed asbestos. Two monitors will be placed in a downwind direction from the exposed asbestos, preferably at the site boundary. One monitor will be placed upwind of the exposed asbestos. If wind direction is unclear, monitors will be placed approximately equidistant around the site. A fifth monitor will serve as a duplicate sampler, and one blank will also be collected.

Three separate sampling events are recommended following a dry period. For the purposes of this study a dry period is defined as five consecutive days without precipitation or frozen snow or water on the surface of the site which could cap any free asbestos. Three separate sampling events will consist of approximately 8 hour shifts and may be conducted over three consecutive days if weather conditions permit. No surface disturbance activities will be performed during the sampling events.

Gilian HFS 513 air pumps fitted with 25mm cellulose ester membrane filters will be used for the monitors. The airflow rate on each pump is variable up to 4 to 5 liters per minute. One to 4 liters per minute will be used on each pump depending on dust conditions on the day of sampling. Pumps have automatic flow controllers and each pump will be calibrated prior to each sampling event with a precision rotameter with a filter cassette in line with the sampling pump and calibration rotameter.

Task 3 - Asbestos Analysis

NIOSH Method 7400 will be used to provide asbestos fiber counts from all samples collected in Subtask 2. In addition, some samples may also be collected and retained for possible future analyses by Transmission Electron Microscopy (TEM) to determine the mineralogy of the asbestos fibers.

NOTE: The range of profiles for the amphiboles will vary up to ± 1 unit for each of the elements present according to the relative detector efficiency of the spectrometer.

c. For an ambiguous structure, obtain EDX spectra on 11 fibers. Label profiles similar to the chrysotile profile as "possible chrysotile." Label profiles similar to the various amphiboles as "possible amphiboles." Label all others as "unknown" or "non-asbestos."

NOTE: Fibers smaller than 0.2 µm in diameter may not produce sufficient peak heights to allow an elemental profile to be determined. Identify these fibers as "unknown".

21. Counting and Sizing:

a. Insert the sample into the specimen grid holder and scan the grid at zero tilt at low magnification (ca. 300 to 500X). Ensure that the carbon film is intact and unbroken over ca. 75% of the grid openings.

b. In order to determine how the grid should be sampled, estimate the number of fibers per grid opening during a low-magnification scan (ca. 1000X). This will allow the analyst to cover most of the area of the grid during the fiber count and analysis. Use the following rules when picking grid openings to count [10]:

(1) Light (<5 fibers per grid opening): count every grid opening in which the carbon film is intact.

(2) Moderate (5 to 25 fibers per grid opening): count every fifth grid opening. If the carbon film is damaged, proceed to the next available grid opening in which the film is intact.

(3) Heavy (>25 fibers per opening): count every tenth grid opening. If the carbon film is damaged, proceed to the next available opening in which the carbon film is intact.

c. Increase magnification to 10,000X. Begin counting at one end of the grid and systematically traverse the grid by rows, reversing direction at row ends. Count at least 2 field blanks per sample set to document possible contamination of the samples. Use the mean fiber count for the field blanks, B, in step 22. Count fibers and asbestos structures using the following rules:

NOTE: Microscopes which traverse nonlinearly or erratically, causing incomplete coverage of the grid opening are not suitable for asbestos analysis [11]. Before using a specific microscope for counting, it should be examined for accuracy and reproducibility of traverses.

(1) Count all particles less than 3 µm diameter which meet the definition of a fiber (aspect ratio $\geq$3:1, with parallel sides).

NOTE: Particles which are of questionable morphology should be analyzed by SAED and EDX to aid in identification.

(2) Size each fiber as it is counted and record the diameter and length (mm):

(a) Move the fiber to the center of the screen. Read the length of the fiber directly from the scale on the screen.

NOTE: For fibers which extend beyond the field of view, the fiber must be moved and superimposed upon the scale until its entire length has been measured.

(b) When a fiber has been sized, return to the starting point and continue the traverse to the next fiber.

(3) Record other asbestos structures according to the following morphological definitions. Label combinations of these structures according to the dominant quality.

(a) Bundle - compact arrangement of parallel fibers in which separate fibers or fibrils may only be visible at the ends or edges of the bundle.

NOTE: Asbestos bundles having aspect ratios of 3:1 or greater and less then 3 µm in diameter are counted as fibers.

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(b) Cluster - network of randomly-oriented interlocking fibers arranged so that no fiber is isolated from the group. Dimensions of clusters can only be roughly estimated and clusters are defined arbitrarily to consist of more than four individual fibers [10].

(c) Matrix - one or more fibers attached to or embedded in a non-asbestos particle.

(4) Count fibers which are partially obscured by the grid.

NOTE: If a fiber is partially obscured by the grid bar at the edge of the field of view, count it as a fiber greater than 5 µm only if more than 2.5 µm of fiber is visible. Otherwise, log the fiber as a "short" fiber, measuring only that portion which is visible.

(5) When counting is complete, calculate the asbestos fiber fraction shorter than 5 µm and thinner than ca. 0.25 µm (the number of asbestos fibers which would be undetected by phase contrast microscopy).

d. Size all fibers using the scale on the fluorescent screen.

NOTE: Data can be recorded directly off the screen in mm and later converted to µm by computer. Count and record identified asbestos fibers and structures >1 µm long of all diameters [11]. However, when comparing TEM data to the PCM counts, all fibers, regardless of identification, greater than 5 µm long and between 0.25 and 3.0 µm diameter which have aspect ratios of 3:1 or greater should be included. This size adjustment is necessary to test comparability of counts between the two methods. If the size-adjusted counts show reasonable equivalency, then further analysis using the small-fiber data will show what fraction of airborne asbestos fibers is being missed by the PCM method.

CALCULATIONS:

22. Calculate and report fiber density on the filter, E, by dividing the total fiber count F, minus the mean field blank count, B, by the number of fields counted, n, (for each sample), and the field area, A_f.

$$E = \frac{\left(\frac{F}{n_f} - \frac{B}{n_b}\right)}{A_f}, \text{ fibers/mm}^2$$

NOTE: The field area, A_f, is considered to be one grid opening, and the size may vary depending upon the type of grids used. This value is known from step 16.

23. Calculate and report the average concentration, C, (fibers/mL) of fibers in the air sample, V (L), using the effective collection area of the filter, A_c (385 mm² for a 25-mm filter):

$$C = \frac{(E)(A_c)}{V \cdot 10^3}$$

24. As an integral part of the report, give the model and manufacturer of the TEM as well as the model and manufacturer of the EDS system.

EVALUATION OF METHOD:

The TEM method has been shown to have a precision of 0.275 (s_r) in an evaluation of mixed amosite and wollastonite fibers. The estimate of the asbestos fraction, however, had a precision of 0.11 (s_r). When this fraction was applied to the PCM count, the overall precision of the combined analysis was 0.20 [3].

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